



TOXICITY OF PYRETHRINS I AND II IN KEROSENE TO THE AMERICAN COCKROACH

By E. R. McGovran and E. L. Mayer, Division of Control Investigations, and Fred Acree, Jr., Division of Insecticide Investigations

The toxicity of pyrethrins I and II to cockroaches has been given some study. Staudinger and Ruzicka (5)1/ tested synthetic pyrethrins I and II on lots of five cockroaches. The pyrethrins were diluted with flour to 1:500, 1:5,000, and 1:25,000. The action of the two pyrethrins was observed to be similar except at 1:5,000, where pyrethrin I killed four of the five roaches treated and pyrethrin II killed only two of the five treated, indicating that pyrethrin I was more effective. The paralytic or knockdown effect of the pyrethrins on the insects was observed by these investigators but they did not record any difference in the two pyrethrins in this respect. Gnadinger and Corl (1) treated cockroaches, Blattella germanica (L.) (Blatta germanica), with alcohol solutions of pyrethrins dispersed as colloidal solutions in water. Twenty-four hours after treatment the result from one concentration of pyrethrin I was recorded as "none dead, all disabled"; in comparison with this, the result from the same concentration of pyrethrin II was recorded as "none dead, all recovered." These investigators concluded that pyrethrin I was slightly more active than pyrethrin II. These results indicated that an investigation of the action of pyrethrins I and II on the American cockroach (Periplaneta, americana (L.)) might be useful in connection with the practical control of this species by the use of pyrethrum sprays.

This investigation also seemed desirable since Sullivan, Haller, McGovran, and Phillips (6) have shown that pyrethrin II was more effective in causing knockdown of houseflies ( $\underline{\text{Musca}}$  domestica (L.)) than pyrethrin I, although the latter caused higher mortalities.

 $<sup>\</sup>underline{1}/\mathrm{The}$  numbers in parentheses refer to the literature cited at the end of this circular.

# Preparation of Pyrethrin Solutions

The pyrethrin concentrates used in the preparation of the kerosene solutions were obtained from petroleum ether oleoresins of Japanese pyrethrum. The separation of the pyrethrins was achieved by partition between acetic acid and petroleum ether as described by LaForge and Haller (3). Further purification of the concentrate high in pyrethrin I was accomplished by distilling it in a molecular still. The pyrethrins were determined by the hydrogenation method of LaForge and Acree (2). Solution 1, in which pyrethrin I predominated, was prepared by dissolving 0.50 gram of a concentrate containing 85 percent of pyrethrin I and 15 percent of pyrethrin II in a highly refined kerosene (Deobase) and diluting the solution to 100 cubic centimeters with the same solvent. Solution 2, which was high in pyrethrin II, was prepared similarly from 0.50 gram of a concentrate containing 95 percent of pyrethrin II and 5 percent of pyrethrin I. Solution 3, containing approximately equal parts of pyrethrins I and II, was prepared as above from 2.0 grams of a concentrate that contained 12.4 percent of pyrethrin I and 12.8 percent of pyrethrin II. These stock solutions, each containing 5 milligrams of total pyrethrins per cubic centimeter, were then diluted with the kerosene to the desired concentration.

## Biological Testing Procedure

After the roaches had been weighed the solutions containing the different concentrations of pyrethrins I and II in refined kerosene were applied with a micropipette, as described by McGovran, Phillips and Mayer (4). The wings of the roaches were held apart so that the liquid was applied to the surface of the dorsal integument upon which it spread rapidly. Each female roach was treated with 0.006 ml. of liquid, and each male with 0.0045 ml. The volume of liquid applied was approximately proportional to the average weight of the individuals and was very close to 0.005 ml. per gram of body weight for both males and females. Three roaches, either all males or all females, were treated in each test. Each test was repeated the number of times shown in tables 1 to 3. After treatment the roaches were released in cages with food and water. The knockdown or paralysis which consisted of the insect being unable to stand or crawl and usually lying on its back with its appendages moving when the cage was shaken, was recorded 15, 30, 45, 60, 90, and 120 minutes after the insecticide was applied. The total of mortality plus knockdown was recorded 24 hours after treatment and the total mortality at the end of 4 days was recorded as shown in tables 1 to 3. The reactions of the male roaches to the insecticides were so different from those of the females that the results were recorded separately.

### Results

The weighing of 322 female roaches in lots of 5 to 8 insects each revealed the average weight of the female roaches to be 1.213 grams. The average weight of the 178 males that were weighed was 0.914 gm.

The data on the knockdown and mortality of the roaches are given in tables 1, 2, and  $\bf 3$ .

Table 1.--Knockdcwn and mortality of female fmerican reach acults caused by two ratios of pyrethrins i sno in herosene

	Mortality	4 cays	Fct.	80	83	77	37	33	7
Knockdown plus	mortality*	24 hours	Pct.	73	53	57	33	20	М
PG	number of minutes	120	Pct,	06	23	83	83	57 E3	Ю
Knockdown in specified		06	Pct.	90	26	83	80	43	13
n spe		09	Fct.	77	24	67	27	37	23
lown		45	Pct. Fct. Fct.	73	92	27	-23	53	100
inockd		30	Pct.	20	23	37	53	20	Ю
124		15	Pct. P	10	63	17	30	23	89
Number	Jo	tests		10	10	10	10	10	10
rins	Potal	content	mg./ml.	1.5	1.5	1.0	1.0	0 0	
Pyrethrins	Retio	H	Pct.	15	92	15	95	C 22 (C)	0
		H	Pct. Pct.	82	Ŋ	22	Ŋ	00 07 07	0
	Series	No.		П	R	B	4	O 01	7

\*This is the number of roaches dead plus the number knocked down 24 hours after treatment.

Table 2. ..-Knockdown and mortality of mair American water caused by two radius of pyrethruns I and II in kerosene

Ma ty	of Cay's	Pot.	83	83	62	42	28	17	0
Knostdown p	24 hours	Pck.	99	69	54	5.7	88,7	17	0
	1.20	Pot.	78	94	75	92	33	20	0
Clf (		Pot.	72	94	75	92	28	20	0
u Ppe	0.50	Pot.	72	94	62	87	800	39	0
ewn i	45 50 -3	Pot. Pot. Pot.	80	94	46	87	28	28	0
Knockdown in specified		Pot.	99	94	37	79	17	11	0
	3	Pot.	17	78	4	28	11	Ŋ	0
Nomber	tests		9	9	oo	ω	9	9	9
Paralining Paralining	content	mg./ml.	0.75	0.75	0.50	0.50	0.25	0.25	0
PANALI		Pot.	15	92	15	92	15	00	0
	ы	Pct.	82	IJ	82	വ	80	Ŋ	0
ON ES	Ma.		7	CZ	Ю	4	ľ	Ø	7

\*This is the number of roaches dead plus the number knocked down 24 hours after treatment.

Table 3.--Knockdown and mortality of American roach adults caused by pyrethrum oleoresin in kerosene

Mortality	4 days	Pct.	76	83	53	88	83	61
Knockdown plus mortality*								
ğ	120	Pct. Pct. Pct. Pct. Pct. Pct.	100	100	29	80	87	72
cifie	06	Pct.	100	100	09	83	87	72
n spe	09	Pct.	26	90	09	29	92	61
own i	45	Pct.	93	77	47	78	75	20
nockd	30	Pct.	80	83	37	61	54	22
X	15	Pct.	09	43	13	44	21	വ
Sex					female		male	
Number			10	10	10	9	œ	9
Total	pyrethrins**	mg./ml.	1.5	1.0	0.5	0.75	0.50	0.25
	No.		1	CV.	Ю	4	വ	9

<sup>\*</sup>This is the number of roaches dead plus the number knocked down 24 hours after treatment.

<sup>\*\*</sup>Prepared from a pyrethrum oleoresin that contained 12.4 percent of pyrethrin I, 12.8 percent of pyrethrin II, and the remainder unidentified.

#### Discussion of Results

The insecticides used in these tests were in all cases mixtures of pyrethrins I and II and other materials. Thus while the effect produced was probably caused mainly by the predominant pyrethrin present, the other pyrethrin which was always present may have influenced the results obtained.

A more rapid knockdown of the roaches occurred with the insecticide rich in pyrethrin II (5 percent pyrethrin I and 95 percent pyrethrin II) than with the material rich in pyrethrin I (85 percent pyrethrin I and 15 percent pyrethrin II) except in series 6, table 2.

The greatest difference in knockdown of the female roaches occurred 15 minutes after treatment, and the difference decreased in the case of the 1.5 mg. per ml. solution until it was not statistically significant after 60 minutes.2/ The differences in knockdown of female roaches caused by the 1.0 mg. per ml. solution were not statistically significant, and only the 15-minute knockdown by the 0.5 mg, per ml. solutions showed a statistically significant difference. The knockdown of male roaches follows the same general pattern, except at the lowest concentration of total pyrethrins (0.25 mg. per ml.) where the knockdown of the insecticide high in pyrethrin II was surprisingly low at first but increased to above that of the insecticide high in pyrethrin I by the 90-minute observation. It appears that at this low concentration the threshold concentration of pyrethrin II that causes the rapid knockdown has not been reached. The knockdown plus mortality at 24 hours showed some decrease from the 2-hour knockdown in all cases except series 4, table 3, and a very large decrease in all instances where the insecticide high in pyrethrin II was used. These latter differences were statistically significant. A few roaches that were treated with the lower concentrations of pyrethrins recovered in the second hour after treatment.

The data presented in tables 1 and 2 record comparisons between the mortalities caused by insecticides high in pyrethrins I and II, 4 days after treatment. These comparisons show that at relatively low mortalities from the practical point of view, namely 77 versus 37 percent and 33 versus 17 percent for females, and 62 versus 42 percent and 28 versus 17 percent for the males, the insecticide that was rich in pyrethrin I caused higher mortalities, although only the first of these differences is statistically highly significant. However, in contrast with the results obtained at these levels of mortality, when sufficient pyrethrum was used to cause mortality of 80 percent or more, the insecticides that were high in pyrethrin I or pyrethrin II gave about equal mortalities 80 versus 83 percent and 83 versus 33 percent on female and male roaches respectively. These data indicate that the comparison of these two insecticides, which was made at relatively low levels of mortality from the practical point of view, was not an accurate comparison of the mortalities these same two insecticides would cause at higher levels of effectiveness, which more nearly approach the mortalities

<sup>2/</sup>The authors wish to express their thanks to Dr. F. M. Wadley for his suggestions on the statistical treatment used.

desired in the practical control of insects. That is, under the conditions of these tests it appears that after a certain concentration was reached the insecticide high in pyrethrin II was just as effective in causing mortality of the roaches as the insecticide high in pyrethrin I.

The insecticide used to obtain the data in table 3 had, in addition to the pyrethrins present, appreciable quantities of pyrethrum oleoresin, as shown in the footnote under table 3. A comparison of the results obtained with this insecticide with the results given in tables 1 and 2 on the basis of total pyrethrins indicates that this insecticide was more effective. The higher toxicity of this insecticide would indicate that the methods of purifying the pyrethrins used in obtaining the data in tables 1 and 2 had reduced the toxicity of the total pyrethrins present somewhat or that the materials in the pyrethrum oleoresins other than pyrethrins I and II had increased the toxicity of the insecticide. These data also show that the female roaches were approximately twice as resistant to this pyrethrum insecticide as the males. The data in tables 1 and 2 also indicate this same relationship in resistance between male and female adult American cockroaches.

A comparison of the 120-minute knockdown, which in most series given in tables 1, 2, and 3 was the highest knockdown observed, with the mortality 4 days after treatment shows that in all but two instances some recovery occurred. In tables 1 and 2 more recovery always occurred with roaches treated with the insecticide high in pyrethrin II than with that high in pyrethrin I. This resulted from two factors; first, in five of the six pairs of series of tests run the pyrethrin II gave the higher knockdown, and in the sixth pair of series the knockdown was equal; and second, in four of the six pairs of series the pyrethrin I caused higher mortalities, in one the mortalities were equal, and in the other a higher mortality was caused by pyrethrin II than by pyrethrin I. The insecticide used in obtaining the data in table 3 is more nearly like the pyrethrum spray that is used commercially to control roaches than are the other two insecticides discussed, but it also shows some recovery of the roaches knocked down in 120 minutes in five of the six series of tests, although the differences are not statistically significant.

Another point of interest is the 100-percent knockdown obtained in series 1 and 2 in table 3. Where 1.5 mg. of total pyrethrins per ml. was used only 3 percent survival occurred, compared with 17 percent survival where 1.0 mg. per ml. was used, indicating that the percentage of mortality among roaches may vary even when a 100-percent knockdown is obtained.

### Summary

Kerosene solutions rich in pyrethrin II (5 percent pyrethrin I, 95 percent pyrethrin II), when applied in measured volumes to adult American cockroaches (Periplaneta americana) with a micropipette, caused more rapid and higher percentages of knockdown than similar insecticides rich in pyrethrin I (85 percent pyrethrin I, 15 percent pyrethrin II) Maximum knockdown was reached in from 30 to 120 minutes.



The insecticide that was rich in pyrethrin I caused higher mortalities at concentrations that caused 17 to 77 percent mortality, but with more concentrated solutions that killed 80 to 83 percent of the roaches the mortalities caused by prethrins I and II preparations were about equal. Female roaches were about twice as resistant to the pyrethrum preparations as male roaches. In 16 of the 18 series of tests made some of the roaches that were knocked down 2 hours after treatment recovered and were living 4 days later.

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